Finding regulatory modules

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CSE527 Computational Biology
Project Presentation

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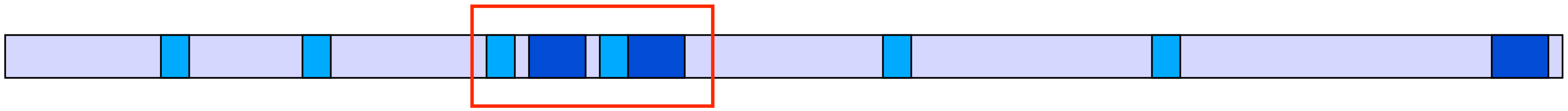
Agenda

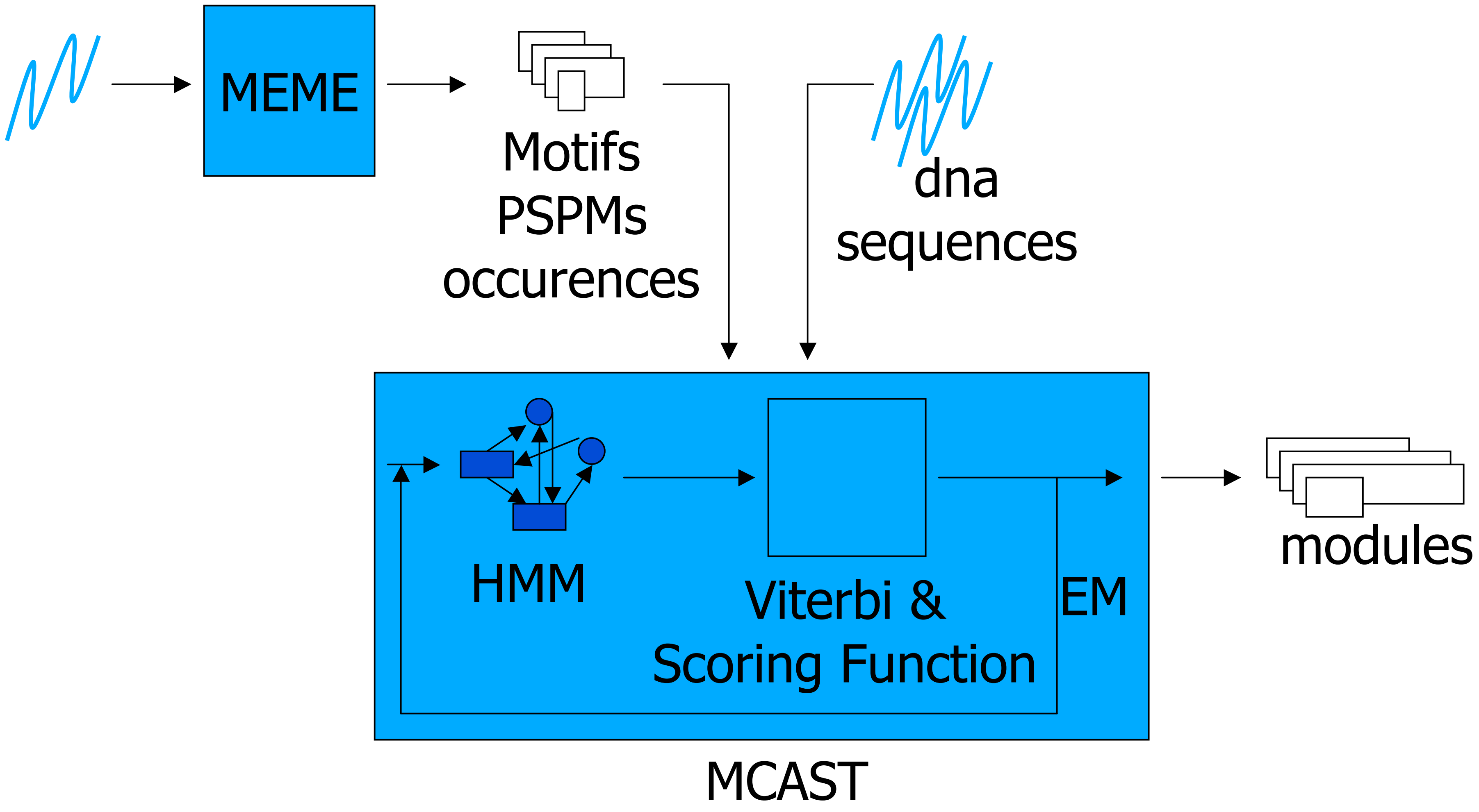
- Importance of module discovery
- MCAST
- My Implementation
- Testing on simulated data
- Testing on real data
- Discussion

Importance of module discovery

- Full understanding of gene functions requires understanding the regulatory machinery
- TFBSs are usually small and appear frequently by chance
- True binding sites appear in clusters



MCAST (part of MetaMEME)



MEME

Motifs
PSPMs
occurences

dna
sequences

HMM

Viterbi &
Scoring Function

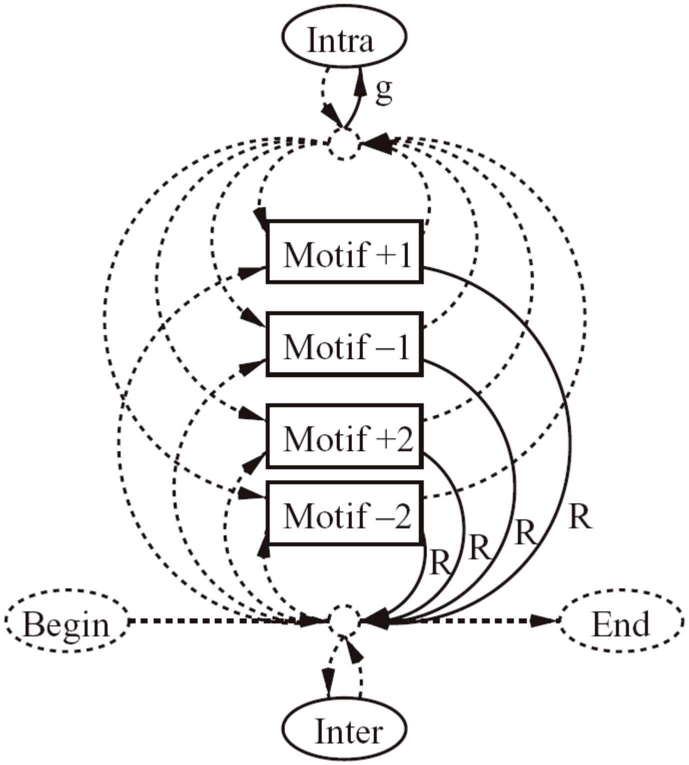
EM

modules

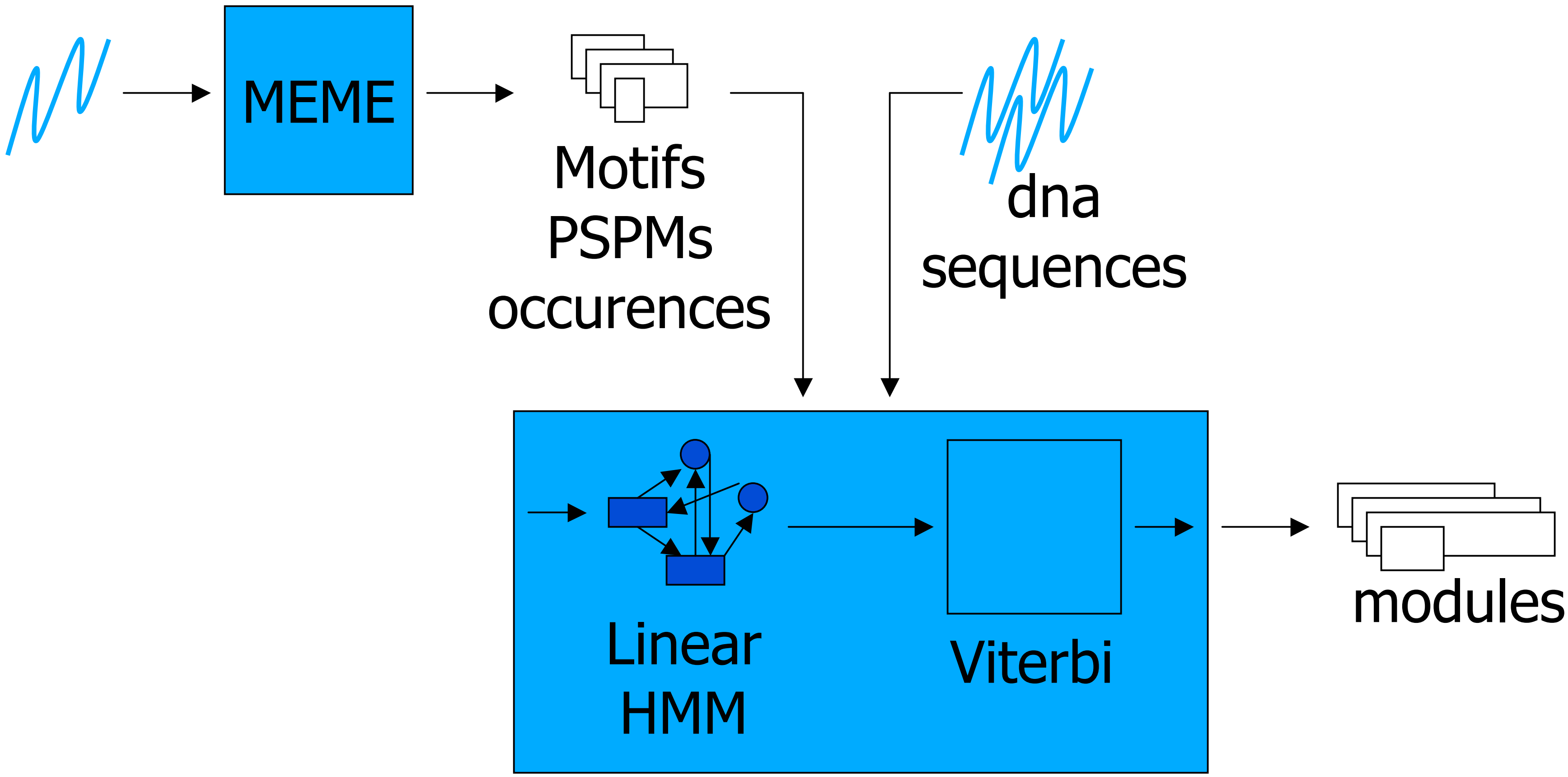
MCAST

MCAST – new ideas

- Sophisticated scoring function
- EM
- HMM

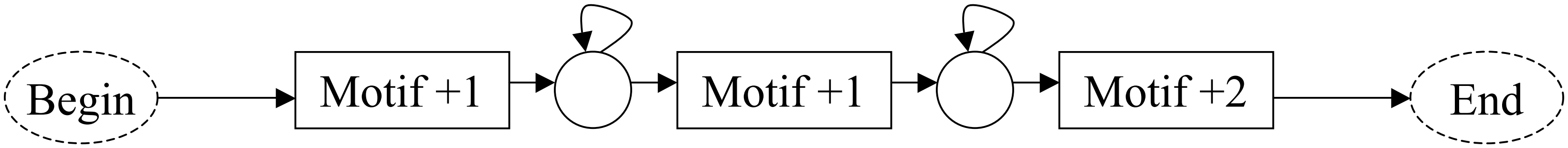


My Implementation



My Implementation

- Written in JAVA
- Includes Random Sequence generator (for simulation data according to HMM)
- Linear HMMs

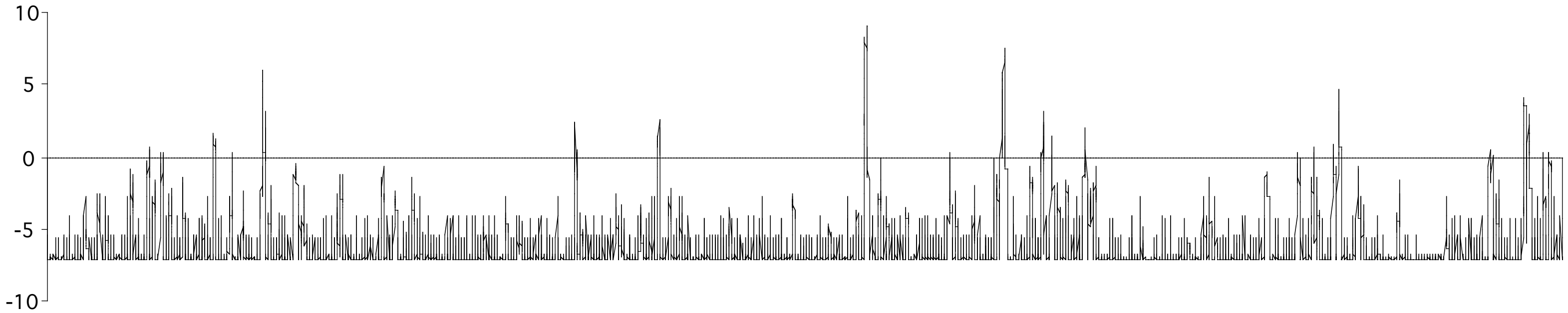


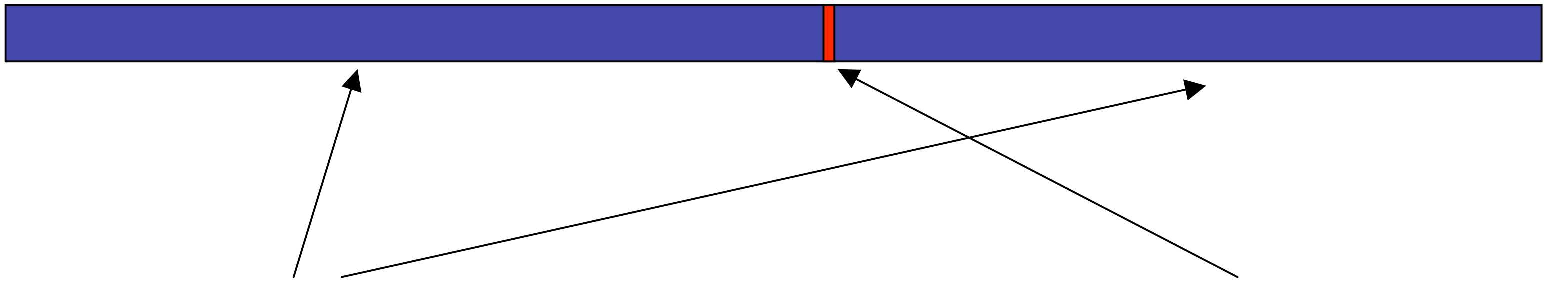
Testing on simulated data

- In real datasets the results depend on a large number of factors, so testing on simulated data first is recommendable.
- I generated a sequence of 16,000 base pairs distributed according to a simple HMM and Drosophila background model (described later)

Testing on simulated data

Log-odds score of sliding window





Background Model

HMM Model

Testing on real data

- Available datasets that contain known modules: Drosophila and human genome
- Motivation: Berman et. al. used a sliding window of 700bp and counted motif occurences in Drosophila sequences. (Successfull though simple)

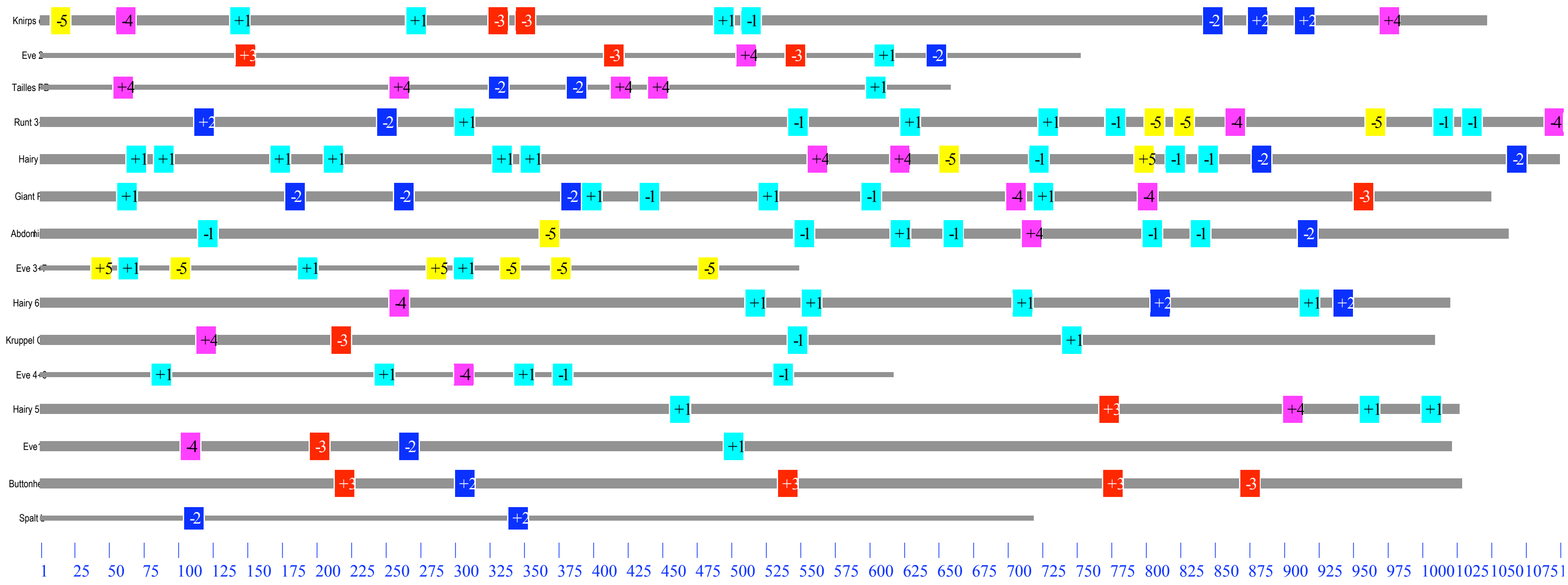
Testing on real data

- Drosophila: ~ 20 modules known upstream of the *even-skipped* gene
- Many contain transcription factor binding sites for Bcd, Cad, Hb, Kr, Kni
- This data has been used by many researchers

Testing on real data

- MEME could not correctly identify the transcription factor binding motifs
- I used PSPMs which were identified by a research team and aligned them to known modules using MAST

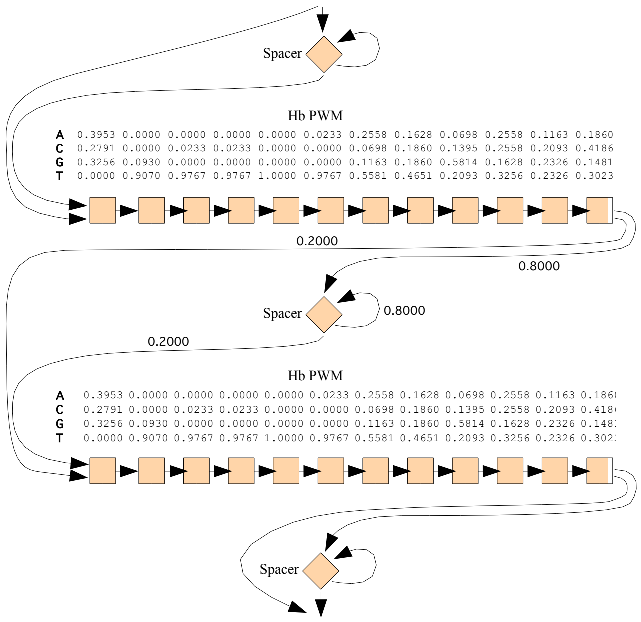
Testing on real data



MAST alignment of known modules
and their transcription factor binding motifs

Testing on real data

- Hardly any common pattern
- But Hb-sites often appear in pairs
- Created a linear HMM



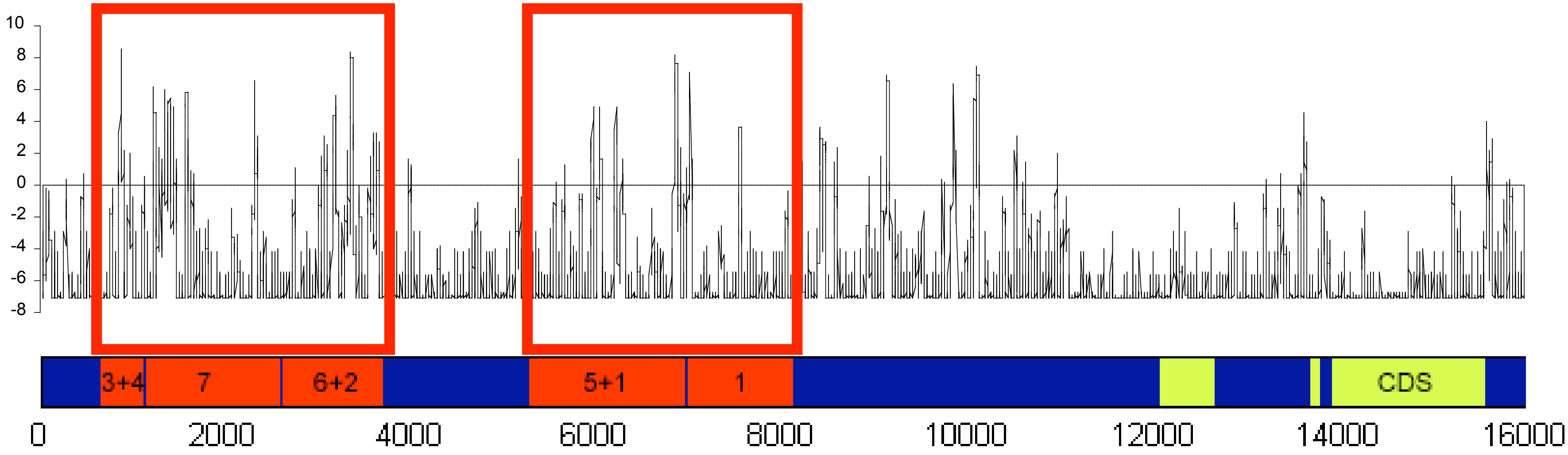
Hb PWM

A	0.3953	0.0000	0.0000	0.0000	0.0000	0.0233	0.2558	0.1628	0.0698	0.2558	0.1163	0.1860
C	0.2791	0.0000	0.0233	0.0233	0.0000	0.0000	0.0698	0.1860	0.1395	0.2558	0.2093	0.4186
G	0.3256	0.0930	0.0000	0.0000	0.0000	0.0000	0.1163	0.1860	0.5814	0.1628	0.2326	0.1481
T	0.0000	0.9070	0.9767	0.9767	1.0000	0.9767	0.5581	0.4651	0.2093	0.3256	0.2326	0.3023

Hb PWM

A	0.3953	0.0000	0.0000	0.0000	0.0000	0.0233	0.2558	0.1628	0.0698	0.2558	0.1163	0.186
C	0.2791	0.0000	0.0233	0.0233	0.0000	0.0000	0.0698	0.1860	0.1395	0.2558	0.2093	0.418
G	0.3256	0.0930	0.0000	0.0000	0.0000	0.0000	0.1163	0.1860	0.5814	0.1628	0.2326	0.148
T	0.0000	0.9070	0.9767	0.9767	1.0000	0.9767	0.5581	0.4651	0.2093	0.3256	0.2326	0.302

Testing on real data



Discussion

Modules not reliably detectable. Why?

- Only regarded motifs on one strand
- Complete Topology or Star Topology HMM instead of Linear HMM
- Geometric distribution of spacer lengths
- Models not accurate
- Log-odds score

Discussion

- Average log-odds scores were higher on the real data than on the simulated data, across the whole sequence. Why?

  Background model which assumes independence might be too simple.

Future Work

- Use EM to train the parameters of the HMM (transition probabilities)